



INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 6 : C07D 47/18, 47/32, 47/16, 47/340, 47/24, 47/300, A61K 31/52		A1	(11) International Publication Number: WO 99/12927 (43) International Publication Date: 18 March 1999 (18.03.99)
(21) International Application Number: PCT/AU98/00748	(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, GH, GM, HR, HU, ID, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG).		
(22) International Filing Date: 11 September 1998 (11.09.98)	Published With international search report.		
(30) Priority Data: PO 9129 11 September 1997 (11.09.97) AU			
(71) Applicant (for all designated States except US): COMMON-WEALTH SCIENTIFIC AND INDUSTRIAL RESEARCH ORGANISATION [AU/AU]; Limestone Avenue, Campbell, ACT 2601 (AU).			
(72) Inventors; and			
(73) Inventors/Applicants (for US only): MARCUCCIO, Sebastian, Mario [AU/AU]; 5 Sporing Court, Endeavour Hills, VIC 3802 (AU). JARVIS, Karen, Elizabeth [AU/AU]; 123 Thompson Crescent, Research, VIC 3095 (AU).			
(74) Agents: SLATTERY, John, M. et al.; Davies Collison Cave, 1 Little Collins Street, Melbourne, VIC 3000 (AU).			
(54) Title: PURINE ACYCLONUCLEOSIDES AS ANTIVIRAL AGENTS			
(57) Abstract			
<p>This invention relates to the use of a compound of formula (1) where: R¹ is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzylxy, NRR' or NRCOR'; R² is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzylxy, NRR' or NRCOR'; and R and R' are independently selected from hydrogen, alkyl and aryl; or a salt and pharmaceutically acceptable derivatives thereof; in the treatment and/or prophylaxis of hepatitis B viral infection.</p>			

FOR THE PURPOSES OF INFORMATION ONLY

Codes used to identify States party to the PCT on the front pages of pamphlets publishing international applications under the PCT.

AL	Albania	ES	Spain	LS	Lesotho	SI	Slovenia
AM	Armenia	FI	Finland	LT	Lithuania	SK	Slovakia
AT	Austria	FR	France	LU	Luxembourg	SN	Senegal
AU	Australia	GA	Gabon	LV	Latvia	SZ	Switzerland
AZ	Azerbaijan	GB	United Kingdom	MC	Monaco	TD	Chad
BI	Bosnia and Herzegovina	GE	Georgia	MD	Republic of Moldova	TG	Togo
BB	Bolivia	GH	Ghana	MG	Madagascar	TJ	Tajikistan
BE	Belgium	GN	Guinea	MK	The former Yugoslav Republic of Macedonia	TM	Turkmenistan
BF	Burkina Faso	GR	Greece	MN	Mali	TR	Turkey
BG	Bulgaria	HU	Hungary	MN	Mongolia	TT	Trinidad and Tobago
BJ	Burkina Faso	IE	Iceland	MR	Mauritania	UA	Ukraine
BR	Brazil	IL	Israel	MW	Malawi	UG	Uganda
BY	Belarus	IS	Iceland	MX	Mexico	US	United States of America
CA	Canada	IT	Italy	NE	Niger	UZ	Uzbekistan
CF	Central African Republic	JP	Japan	NL	Netherlands	VN	Viet Nam
CG	Congo	KE	Kenya	NO	Norway	YU	Yugoslavia
CH	Switzerland	KG	Kyrgyzstan	NZ	New Zealand	ZW	Zimbabwe
CI	Côte d'Ivoire	KP	Democratic People's Republic of Korea	PL	Poland		
CM	Cameroon	KR	Republic of Korea	PT	Portugal		
CN	China	KZ	Kazakhstan	RO	Romania		
CU	Cuba	LC	Saint Lucia	RU	Russian Federation		
CZ	Czech Republic	LI	Liechtenstein	SD	Sudan		
DE	Germany	LK	Sri Lanka	SE	Sweden		
DK	Denmark	LR	Liberia	SG	Singapore		

- 1 -

ANTIVIRAL AGENTS

The present invention relates to the use of purine acyclonucleosides as agents in the treatment and/or prophylaxis of hepatitis B, pharmaceutical compositions for use in such therapy and novel purine acyclonucleosides.

5

Infection with human hepatitis B virus is a major public health problem because of the ability of the virus to cause acute and chronic infections. Chronic hepatitis B virus infection (hereinafter referred to as HBV) causes serious liver disease in humans and frequently results in cirrhosis and hepatocellular carcinoma. Currently there is no 10 effective therapy for the successful management of chronic HBV infections. The > 250 million chronic HBV carriers throughout the world are unable to benefit from the commercial vaccine now available.

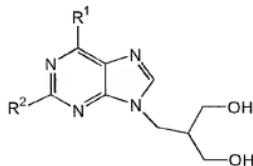
15 Currently available therapies for HBV provide inadequate levels of efficacy or are accompanied by deleterious side effects. Accordingly, a need exists for effective treatments for HBV.

It has now been discovered that compounds of formula (1) are active agents against hepatitis B virus.

20

Accordingly, in one aspect of the present invention there is provided the use of a compound of formula (1)

25



(1)

- 2 -

where:

R¹ is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or NRCOR';

R² is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or NRCOR'; and

R and R' are independently selected from hydrogen, alkyl and aryl;

or salts or pharmaceutically acceptable derivatives thereof;

in the treatment and/or prophylaxis of hepatitis B viral infection.

10 These compounds have been found to exhibit surprisingly good activity in an anti-hepatitis B assay.

Preferably R¹ is hydroxy or a group capable of being converted *in vivo* to hydroxy.

Preferably R² is amino; or a group which is capable of being converted *in vivo* to amino.

15 The invention further provides a method for the treatment or prophylaxis of hepatitis B viral infection which method includes administering to a patient in need thereof an effective amount of a compound of formula (1), its salts, and pharmaceutically acceptable derivatives.

The present invention also provides a compound of formula (1), its salts, and
20 pharmaceutically acceptable derivatives for use in treatment or prophylaxis of HBV.

The compounds of the invention may further be used in the manufacture of a medicament for the treatment or prophylaxis of HBV. Accordingly, the present invention provides pharmaceutical compositions for said treatment or prophylaxis which include a compound of formula (1), its salts or pharmaceutically acceptable derivatives in
25 association with a pharmaceutically acceptable carrier or diluent.

- 3 -

The present invention also provides the use of a compound of formula (1), its salts or pharmaceutically acceptable salts thereof in the manufacture of a medicament for use in the treatment or prophylaxis of HBV.

The salts of the compounds of formula (1) are preferably pharmaceutically acceptable, but it will be appreciated that non-pharmaceutically acceptable salts also fall within the scope of the present invention, since these are useful as intermediates in the preparation of pharmaceutically acceptable salts. The pharmaceutically acceptable salts may include conventional non-toxic salts or quaternary ammonium salts of these compounds, which may be formed for example from organic or inorganic acids or bases.

5 Examples of such acid addition salts include, but are not limited to, those formed with pharmaceutically acceptable acids such as acetic, propionic, citric, lactic, methanesulphonic, toluenesulphonic, benzenesulphonic, salicylic, ascorbic, hydrochloric, orthophosphoric, sulphuric and hydrobromic acids. Base salts includes, but is not limited to, those formed with pharmaceutically acceptable cations, such as sodium, potassium,

10 lithium, calcium magnesium, ammonium and alkylammonium. They may be formed by treating a compound of formula (1) with an appropriate metal hydroxide. Also, basic nitrogen-containing groups may be quaternised with such agents as lower alkyl halides, such as methyl, ethyl, propyl, and butyl chlorides, bromides and iodides; dialkyl sulfates like dimethyl and diethyl sulfate; and others.

15

20 The compounds of the invention may be in crystalline form or as solvates (e.g. hydrates) and it is intended that both forms are within the scope of the present invention. Methods of solvation are generally known within the art.

25 Pharmaceutically acceptable derivatives may include any pharmaceutically acceptable salt, hydrate, prodrug, or any other compound which, upon administration to a subject, is capable of providing (directly or indirectly) a compound of formula (1) or an antivirally active metabolite or residue thereof. For example, compounds where a hydroxy group on the acyclic sidechain has been replaced with a phosphate ester are within the scope of pharmaceutically acceptable derivatives.

The term "prodrug" is used in its broadest sense and encompasses those derivatives

that are converted *in vivo* to the compounds of the invention. Such derivatives would readily occur to those skilled in the art, and include, for example, compounds where R¹ is a group which is capable of being converted *in vivo* to hydroxy; or R² is a group which is capable of being converted *in vivo* to amino; or compounds where a free hydroxy group 5 on the acyclic sidechain is converted into a group, for example an ester, a carbonate or a carbamate, which is capable of being converted *in vivo* back to a hydroxy group. A prodrug may include modifications to one or more of the functional groups of a compound of the invention.

Throughout this specification the phrase "a group which is capable of being 10 converted *in vivo*" used in relation to another functional group includes all those functional groups or derivatives of such groups which upon administration into a mammal may be converted into the stated functional group. Those skilled in the art may readily determine whether a group may be capable of being converted *in vivo* into the stated functional group using routine enzymatic or animal studies.

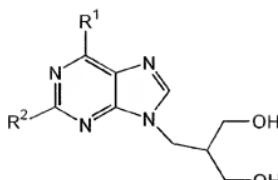
15 It will be appreciated that some derivatives of compounds of formula (1) may have an asymmetric centre, and therefore are capable of existing in more than one stereoisomeric form. The invention extends to each of these forms individually and to mixtures thereof, including racemates. The isomers may be separated conventionally by chromatographic methods or using a resolving agent. Alternatively, the individual 20 isomers may be prepared by asymmetric synthesis using chiral intermediates, or enzymes.

Preferably R¹ is hydroxy or a group which is capable of being converted *in vivo* to hydroxy.

Preferably R² is amino, or a group which is capable of being converted *in vivo* to amino.

25 Some of the compounds of formula (1) are novel and accordingly the invention also provides compounds of formula (1a):

- 5 -



(1a)

where:

R¹ is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or NRCOR';

10 R² is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or NRCOR'; and

R and R' are independently selected from hydrogen, alkyl and aryl;

and salts and pharmaceutically acceptable derivatives thereof;

provided that the following compounds are excluded;

15 9-[3-Hydroxy-2-hydroxymethylprop-1-yl]-guanine, 9-[3-Hydroxy-2-hydroxymethylprop-1-yl]-adenine, 9-[(2-isopropyl-1,3-dioxan-5-yl)methyl]-guanine, and 9-[(2-isopropyl-1,3-dioxan-5-yl)methyl]-adenine.

The compound of formula (1) where R¹ is OH and R² is NH₂ has been reported by Martin *et al.* (1986) as being inactive against herpes simplex virus type 1 and as a poor 20 substrate for the thymidine kinase of that virus.

Throughout this specification the term alkyl, used either alone or in compound words such as haloalkyl or alkyl acids is denoted, unless otherwise defined, to mean both the straight chain C₁₋₃₀alkyl or branched chain C₃₋₃₀alkyl and the branched or unbranched C₃₋₃₀cycloalkyl. Unless otherwise defined, such groups may be saturated or 25 unsaturated.

- 6 -

The term "aryl" as used herein refers to any compound which includes or consists of one or more aromatic rings. The aromatic rings may be carbocyclic, heterocyclic or pseudoaromatic, and may be mono or polycyclic ring systems and preferably have 2 to 20 carbon atoms. The aromatic rings may also have one or more heteroatoms selected from 5 N, S, O and P. Examples of suitable rings include but are not limited to benzene, biphenyl, terphenyl, quaterphenyl, naphthalene, tetradyronaphthalene, 1-benzylnaphthalene, anthracene, dihydroanthracene, benzanthracene, dibenzanthracene, phenanthracene, perylene, pyridine, 4-phenylpyridine, 3-phenylpyridine, thiophene, benzothiophene, naphthothiophene, thianthrene, furan, pyrene, isobenzofuran, 10 chromene, xanthene, phenoxathiin, pyrrole, imidazole, pyrazole, pyrazine, pyrimidine, pyridazine, indole, indolizine, isoindole, purine, quinoline, isoquinoline, phthalazine, quinoxaline, quinazoline, pteridine, carbazole, carboline, phenanthridine, acridine, phenanthroline, phenazine, isothiazole, isooxazole, phenoxyazine and the like, each of which may be optionally substituted. The term "pseudoaromatic" refers to a ring system 15 which is not strictly aromatic, but which is stabilized by means of delocalization of electrons and behaves in a similar manner to aromatic rings. Examples of pseudoaromatic rings include but are not limited to furan, thiophene, pyrrole and the like.

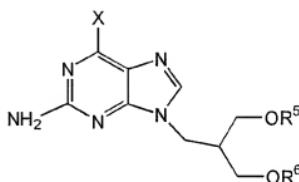
Throughout this specification the term alkoxy, used either alone or in compound words such as haloalkoxy is denoted, unless otherwise defined, to mean both the straight 20 chain C_{1-30} alkoxy or branched chain C_{3-30} alkoxy and the branched or unbranched C_3 ,
₃₀cycloalkoxy. Unless otherwise defined, such groups may be saturated or unsaturated.

Unless otherwise stated the term "ester" is denoted to mean those compounds or derivatives which correspond to the ester formed by reaction of an alcohol with an organic acid, preferably a carboxylic acid. Particularly preferred carboxylic acids from which 25 esters may be formed include amino acids and alkyl acids. Preferred amino acids are aliphatic amino acids such as valine and isoleucine, preferably in the L-form. Preferred alkyl acids include C_2-C_4 alkyl acids, and fatty acids from $C_{11}-C_{22}$ such as lauryl, myristoyl, palmitoyl, stearoyl, eicasanoyl, behenoyl, myristoleic, myristelaidic, palmitoleic, palmitelaidic, n6-octadecenoic, oleic, elaidic, erucic or brassidic acids.

- 7 -

A preferred group of compounds of formula (1) and formula (1a) and pharmaceutically acceptable derivatives of the compounds of formula (1) and formula (1a) are those compounds of formula (4)

5



(4)

10 where:

X is hydrogen or hydroxy;

R⁵ and R⁶ are the same or different and together with the oxygen atom to which they are attached form a hydroxy group, an ester, a carbonate, a carbamate, or a thiocarbonate; preferably a hydroxy or an ester;

15 and salts thereof.

Examples of some compounds of formula (4) are shown in Table 1.

Table 1

20
25

X	R ⁵	R ⁶
H	H	H
OH	H	H
H	acetyl	acetyl
OH	acetyl	acetyl
H	acetyl	H
OH	acetyl	H
H	valyl	valyl

- 8 -

	OH	valyl	valyl
	H	valyl	H
	OH	valyl	H
	H	valyl	acetyl
5	OH	valyl	acetyl
	H	isoleucyl	isoleucyl
	OH	isoleucyl	isoleucyl
	H	isoleucyl	acetyl
	OH	isoleucyl	acetyl
10	H	valyl	stearyl
	OH	valyl	stearyl
	H	valyl	octadecenyl
	OH	valyl	octadecenyl
	H	isoleucyl	stearyl
15	OH	isoleucyl	stearyl
	H	isoleucyl	octadecenyl
	OH	isoleucyl	octadecenyl
	H	valyl	palmityl
	OH	valyl	palmityl
20	H	valyl	elaidyl
	OH	valyl	elaidyl
	H	isoleucyl	palmityl
	OH	isoleucyl	palmityl
	H	isoleucyl	elaidyl
25	OH	isoleucyl	elaidyl
	H	H	stearyl
	OH	H	stearyl
	H	H	octadecenyl
	OH	H	octadecenyl
30	H	H	palmityl
	OH	H	palmityl
	H	H	elaidyl
	OH	H	elaidyl
	H	H	cholyl
35	OH	H	cholyl
	H	valyl	cholyl

- 9 -

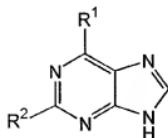
	OH	valyl	cholyl
	H	propionyl	propionyl
	OH	propionyl	propionyl
	H	isopropionyl	isopropionyl
5	OH	isopropionyl	isopropionyl
	H	H	butyryl
	OH	H	butyryl
	H	butyryl	butyryl
	OH	butyryl	butyryl

10

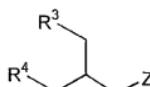
In yet another aspect of the invention there is provided a method for the manufacture of the compounds of formula (1), their salts and pharmaceutically acceptable derivatives. The compounds may be prepared by reacting a purine derivative (2) with a compound of formula (3). The group R¹ of purine derivative (2) may be any group listed under R¹ for 15 the compound of formula (1) or any group that may be converted by methods known in the art to such groups, such groups include chlorine, bromine or iodine; and R² may be any group listed under R² for the compound of formula (1) or any group that may be converted by methods known in the art to such groups. In compound (3) Z is any suitable leaving group, such as methane sulfonate or bromine, and R³ and R⁴ are hydroxy or a 20 group that may be converted into hydroxy, such groups include ethers and esters.

Methods for such conversion are known to those skilled in the art. R³ and R⁴ may be joined together to form an optionally substituted 5 or 6 membered ring system.

25



(2)



(3)

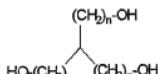
Compounds of formula (2) are commercially available or may be prepared by literature

- 10 -

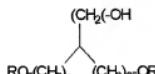
procedures. Compounds of formula (3) may be prepared by literature procedures or along the lines of the procedures described in Steps A to C of Example 1, Steps A to E of the alternative route to Example 1, see Schemes 1 and 2 respectively. Those in the art will appreciate that a number of variations may be made to the methodology actually 5 exemplified.

The procedure for manufacture of the compound of formula (3) outlined in Example 1 Scheme 1 is particularly useful and may be more broadly applicable to the synthesis of acyclic nucleoside analogues than is outlined in the specific example. The general procedure of Step B forms a further aspect of the present invention. Accordingly, 10 a symmetrical triol (5), preferably where n is 1 or 2, may be converted into a diester (6) by treatment with about one equivalent of a trialkylorthoester, preferably triethylorthoacetate, under appropriate conditions, followed by treatment with about one equivalent of water under appropriate conditions, followed by treatment with water under appropriate conditions. The diester may then be isolated by conventional procedures, 15 conveniently this is done via a careful neutralisation of the mixture with a mild base, for example sodium bicarbonate, followed by extraction into an organic solvent. Those skilled in the art may readily determine which conditions are appropriate in view of the particular triol and trialkylorthoester selected.

20



(5)



-OR = an ester

(6)

The remaining hydroxyl group on the diester (6) may be converted into a leaving 25 group, Z, to give diester compounds, which where n is 1 are of general formula (3).

- 11 -

Compounds of formula (6), such as 2-hydroxymethyl-1,3-propanediol diacetate, and diester compounds including those of general formula (3), such as 3-acetoxy-2-acetoxymethylprop-1-yl methanesulfonate, are novel and form a further aspect of the invention. Preferably n is 1 in compound (5) and (6).

5 In accordance with conventional processes known in the art, the acyclic hydroxyl groups of compounds of formula (1) may be readily converted into esters, ethers or phosphate groups or a mixture of these groups on the acyclic chain. In some instances it may be useful to utilise protected intermediates of the compound of formula (1) in order to prepare the desired final derivative. Such intermediates may be prepared in accordance
10 with standard procedures and when no longer required the protecting groups removed using standard procedures, such as those described by Greene. Examples of suitable protecting groups are trimethylsilyl and monomethoxytrityl groups.

Acylation and alkylation may be carried out using any conventional procedure such as those generally known in the art or described or referenced in the Third Edition of
15 March's Advanced Organic Chemistry published by Wiley-Interscience. Examples of acylating agents suitable for the process of acylating the compounds of formula (1) are carboxylic acids, acid halides and acid anhydrides. The reaction may be carried out in a conventional manner, for example in a solvent such as pyridine, dimethylformamide, etc., optionally in the presence of a coupling agent such as N,N'-dicyclohexylcarbodiimide, and
20 optionally in the presence of a catalytic base such as 4-dimethylaminopyridine. The product of the reaction may be isolated in a conventional manner. Examples of alkylating agents suitable for the process of alkylating compounds of formula (1) are alkyl halides, such as methyl, ethyl, propyl, and benzyl chlorides, bromides and iodides; and dialkyl sulfates like dimethyl and diethyl sulfate.

25 In the case of amino acids or their functional equivalents, for example acid halides, it may be advantageous, in order to avoid side reactions, to use amino protected derivatives of the amino acid or amino acid equivalent, for example benzyloxycarbonyl derivatives. Such derivatives are commercially available. The protecting groups may be removed utilising standard procedures.

- 12 -

The acylation or alkylation reactions may produce a single derivative of compound (1), incorporating one or more acyl or alkyl groups, or may produce a mixture of compounds incorporating acyl or alkyl groups. The outcome depends on a number of factors, such as the relative amounts and chemical nature of the reactants, the physical 5 conditions of the reaction, and the solvent system. Any mixture produced in this way may be separated using standard techniques, preferably chromatography.

It will be appreciated by one skilled in the art that it is possible to produce derivatives of compounds of formula (1) that may have a mixture of different acyl and/or alkyl groups. Such derivatives are within the scope of the present invention.

10 Protected intermediates of the compounds of formula (1) may also be used to prepare derivatives of compound (1) incorporating phosphate esters.

The compounds of this invention may also be useful in combination with known antiviral or antiretroviral agents or other pharmaceuticals used in the treatment of viral infections. Representative examples of these additional pharmaceuticals include 15 immunomodulators, immunostimulants, and antibiotics. Exemplative anti-viral agents include AZT, 3TC, acyclovir, famciclovir, ddI, ddC, ganciclovir, saquinavir, lioviride, other non-nucleotide reverse transcriptase (RT) inhibitors and protease inhibitors. Exemplative immunomodulators and immunostimulants include various interleukins, cytokines, antibody preparations, blood transfusions and cell transfusions. Exemplative 20 antibiotics includes antifungal agents, antibacterial agents and anti-Pneumocystis carinii agents.

If formulated as a fixed dose, such combination products employ the compounds of this invention in the dosage ranges described below and the other pharmaceutically active agent within its approved dosage range. The compounds of the invention may be used 25 sequentially with known anti-viral, anti-retroviral or pharmaceutical agents when a combination formulation is inappropriate.

By an effective amount is meant a quantity of active compound which will upon single or multiple dose administration to the patient be effective in controlling the viral

infections, such as HBV, or in achieving a blood or tissue level in the patient that corresponds to a concentration of the active compound that has been shown to inhibit a virus, such as HBV, in an assay known to predict for clinical anti-viral activity of chemical compounds. For example the assay described by Korba and Gerin.

5 As used herein the term controlling the viral infections refers to slowing, interrupting, arresting or stopping its growth or replication and does not necessarily indicate a total elimination of the virus.

Controlling the viral infections will be useful in the treatment and/or prophylaxis of such viral infections.

10 When a compound of the invention is administered to a human subject the daily dosage can normally be determined by the attending physician with the dosage generally varying according to the age, weight, and response of the individual patient, as well as the severity of the patient's symptoms. In general a suitable dose of the compound of the invention will be in the range of 0.1 to 50 mg per kilogram body weight of the recipient
15 per day, preferably in the range of 0.5 to 10 mg per kilogram body weight per day. The desired dose is preferably presented as two, three, four, five, six or more sub-doses administered at appropriate intervals throughout the day. These sub-doses may be administered in unit dosage forms, for example, containing 1 to 1000 mg, preferably 10 to 500 mg of active ingredient per unit dosage form.

20 The compounds according to the invention, also referred to herein as the active ingredient, may be administered for therapy by any suitable route, including oral, rectal, nasal, topical (including buccal and sublingual), vaginal and parenteral (including subcutaneous, intramuscular, intravenous and intradermal). Preferably, administration will be by the oral route, however it will be appreciated that the preferred route will vary
25 with the condition and age of the recipient, the nature of the invention and the chosen active ingredient. When administered by the oral route a prodrug of the active compound which is more efficiently absorbed than the unmodified compound is generally preferred.

The compositions of the present invention comprise the compound of formula (1).

- 14 -

optionally as a salt or other pharmaceutically acceptable derivative, together with one or more pharmaceutically acceptable carriers, diluents or excipients therefor, and optionally other therapeutic agents. Each carrier, diluent or excipient must be pharmaceutically "acceptable" in the sense of being compatible with the other ingredients of the composition 5 and not injurious to the patient. Compositions include those suitable for oral, rectal, nasal, topical (including buccal and sublingual), vaginal or parenteral (including subcutaneous, intramuscular, intravenous and intradermal) administration. The compositions may conveniently be presented in unit dosage form and may be prepared by methods well known in the art of pharmacy. Such methods include the step of bringing 10 into association the active ingredient with the carrier, diluent or excipient which includes one or more accessory ingredients. In general, the compositions are prepared by uniformly and intimately bringing into association the active ingredient with liquid carriers or finely divided solid carriers or both, and then if necessary shaping the product.

Compositions of the present invention suitable for oral administration may be 15 presented as discrete units such as capsules, sachets or tablets each containing a predetermined amount of the active ingredient; as a powder or granules; as a solution or a suspension in an aqueous or non-aqueous liquid; or as an oil-in-water liquid emulsion or a water-in-oil liquid emulsion. The active ingredient may also be presented as a bolus, electuary or paste.

20 Tablets may be made by compression or moulding, optionally with one or more accessory ingredients. Compressed tablets may be prepared by compressing in a suitable machine the active ingredient in a free-flowing form such as a powder or granules, optionally mixed with a binder (e.g. inert diluent, preservative disintegrant (e.g. sodium starch glycollate, cross-linked povidone, cross-linked sodium carboxymethyl cellulose) 25 surface-active or dispersing agent. Moulded tablets may be made by moulding in a suitable machine a mixture of the powdered compound moistened with an inert liquid diluent. The tablets may optionally be coated or scored and may be formulated so as to provide slow or controlled release of the active ingredient therein using, for example, hydroxypropylmethyl cellulose in varying proportions to provide the desired release 30 profile. Tablets may optionally be provided with an enteric coating, to provide release in

parts of the gut other than the stomach.

Compositions suitable for topical administration in the mouth include lozenges comprising the active ingredient in a flavoured base, usually sucrose and acacia or tragacanth gum; pastilles comprising the active ingredient in an inert base such as gelatin 5 and glycerin, or sucrose and acacia gum; and mouthwashes comprising the active ingredient in a suitable liquid carrier.

Compositions for rectal administration may be presented as a suppository with a suitable base comprising, for example, cocoa butter.

Compositions suitable for vaginal administration may be presented as pessaries, 10 tampons, creams, gels, pastes, foams or spray formulations containing in addition to the active ingredient such carriers as are known in the art to be appropriate.

Compositions suitable for parenteral administration include aqueous and non-aqueous isotonic sterile injection solutions which may contain anti-oxidants, buffers, bacteriostats and solutes which render the composition isotonic with the blood of the 15 intended recipient; and aqueous and non-aqueous sterile suspensions which may include suspending agents and thickening agents. The compositions may be presented in unit-dose or multi-dose sealed containers, for example, ampoules and vials, and may be stored in a freeze-dried (lyophilised) condition requiring only the addition of the sterile liquid carrier, for example water for injections, immediately prior to use. Extemporaneous injection 20 solutions and suspensions may be prepared from sterile powders, granules and tablets of the kind previously described.

Preferred unit dosage compositions are those containing a daily dose or unit, daily sub-dose, as herein above recited, or an appropriate fraction thereof, of an active ingredient.

25 The compounds according to the invention may also be presented for use in the form of veterinary compositions, which may be prepared, for example, by methods that are conventional in the art. Examples of such veterinary compositions include those adapted for:

- 16 -

- (a) oral administration, external application, for example drenches (e.g., aqueous or non-aqueous solutions or suspensions); tablets or boluses; powders, granules or pellets for admixture with feed stuffs; pastes for application to the tongue;
- 5 (b) parenteral administration for example by subcutaneous, intramuscular or intravenous injection, e.g. as a sterile solution or suspension;
- (c) topical application, e.g. as a cream, ointment or spray applied to the skin; or
- (d) intravaginally, e.g. as a pessary, cream or foam.

10 It should be understood that in addition to the ingredients particularly mentioned above, the compositions of this invention may include other agents conventional in the art having regard to the type of composition in question, for example, those suitable for oral administration may include such further agents as sweeteners, thickeners and flavouring agents.

15

EXAMPLES

Examples are provided to assist in the further understanding of the invention. Particular materials, and conditions employed are intended to be illustrative of the invention and not limitative of the reasonable scope thereof.

20

2-Amino-6-chloropurine was obtained commercially in 98% purity from Chugai Boyeki Co., Ltd. 2-Amino-6-chloropurine was converted to 2-amino-6-iodopurine according to the method of Bisacchi *et al.* Unreferenced reagents were obtained commercially and used as supplied, unless otherwise specified.

25

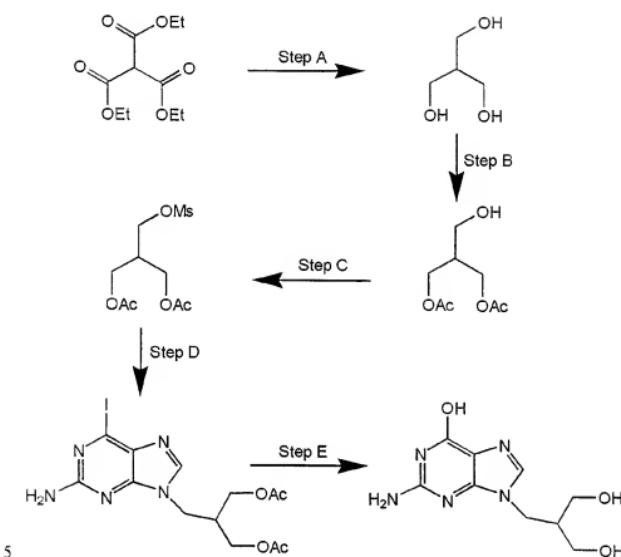
All temperatures are given in degrees Celsius.

- 17 -

Example 1

Scheme 1

Reaction sequence described in Example 1



- 18 -

Step A

2-Hydroxymethyl-1,3-propanediol

5 A modified procedure of Harnden *et al* was used. To a solution of borane-methylsulfide complex (10M) (14 ml, 0.14 mol) in toluene (65 ml) under a nitrogen atmosphere, at gentle reflux, was added dropwise triethylmethane tricarboxylate (9.68 g, 0.0417 mol). The reaction mixture was refluxed for 7.5 hr with distillation of dimethyl sulfide. The reaction mixture was cooled to room temperature and 10 methanol (40 ml) was added dropwise. The reaction mixture was stirred at room temperature overnight, then the solvents were removed and the residue co-evaporated with methanol repetitively. The residue was chromatographed on silica with 25% methanol in dichloromethane to afford 2-hydroxymethyl-1,3-propanediol as a pale lemon oil (3.18 g, 72%). ^1H n.m.r. (DMSO-d₆) 1.60, septet, J = 15 5.5Hz, 1H; 3.40, t, J = 5.5Hz, 2H; 4.31, t, J = 5.5Hz, 3H.

Step B

2-Hydroxymethyl-1,3-propanediol diacetate

20 To a solution of 2-hydroxymethyl-1,3-propanediol (3.18 g, 0.03 mol), trifluoroacetic acid (1.54 ml, 0.02 mol) in N,N-dimethylformamide (35 ml) under a nitrogen atmosphere, was added triethylorthoacetate (6.09 ml, 0.033 mol). The reaction mixture was stirred for 1.5 hr then water (0.63 ml, 0.035 mol) was added. 25 The reaction mixture was stirred for a further 1.5 hr then triethylorthoacetate (6.26 ml, 0.034 mol) was added. The reaction mixture was stirred for a further 1.5 hr

- 19 -

then water (1.74 ml, 0.097 mol) was added. After 1 hr sodium bicarbonate was carefully added until the reaction mixture was neutralized. Water was added and the solution was extracted with dichloromethane (3x). The combined extracts were washed with water, dried over magnesium sulfate and concentrated to afford 2-5 hydroxymethyl-1,3-propanediol diacetate as a colourless oil (4.50 g, 79%). ¹H n.m.r. (CDCl₃) 2.06, s, 6H; 2.18, septet, *J* = 6Hz, 1H, 2.54, br s, 1H, 3.62, d, *J* = 6Hz, 2H; 4.15, d, *J* = 6Hz, 4H.

Step C

10

3-Acetoxy-2-acetoxymethylprop-1-yl methanesulfonate

A solution of 2-hydroxymethyl-1,3-propanediol diacetate (4.49 g, 0.0236 mol) in dichloromethane (40 ml) under a nitrogen atmosphere, was cooled to -5°. 15 Triethylamine (4.93 ml, 0.0354 mol) was added, followed by dropwise addition of a solution of methanesulfonyl chloride (2.19 ml, 0.0283 mol) in dichloromethane (20 ml). The reaction was stirred a further 2 hr at 0° then warmed to room temperature. The reaction mixture was washed with 1.5M hydrochloric acid (3 x 30 ml), saturated sodium bicarbonate solution (30 ml) and brine (30 ml), dried 20 over magnesium sulfate and concentrated to afford 3-acetoxy-2-acetoxymethylprop-1-yl methanesulfonate as a pale brown oil (5.02 g, 79%). ¹H n.m.r. (CDCl₃) 2.0, s, 6H; 2.48, septet, *J* = 6Hz, 1H; 3.03, s, 3H; 4.07 - 4.24, m, 4H; 4.29, d, *J* = 6Hz, 2H.

25 Step D

9-[3-Acetoxy-2-acetoxymethylprop-1-yl]-2-amino-6-iodopurine

- 20 -

A mixture of 3-acetoxy-2-acetoxymethylprop-1-yl methanesulfonate (5.02 g, 0.0187 mol), 2-amino-6-iodopurine (4.66 g, 0.0178 mol) and potassium carbonate (7.38 g, 0.0534 mol) in N,N-dimethylformamide (250 ml) under a nitrogen atmosphere, was stirred at 60° for 2 days. The N,N-dimethylformamide was removed *in vacuo*, water was added and the residue was extracted with ethyl acetate (3 x 100 ml). The extracts were washed with water and brine, dried over magnesium sulfate, then filtered through a plug of silica, washing thoroughly with ethyl acetate. The filtrate was concentrated to give the crude product as a yellow solid (5.96 g). This was recrystallised from methanol and diethyl ether to afford 9-10 [3-acetoxy-2-acetoxymethylprop-1-yl]-2-amino-6-iodopurine as pale lemon crystals (4.43 g, 55%). ¹H n.m.r. (DMSO-d₆) 1.95, s, 6H; 2.56 - 2.78, m, 1H; 3.90 - 4.10, m, 4H; 4.12, d, *J* = 7Hz, 2H; 6.83, s, 2H; 8.09, s, 1H.

Step E

15

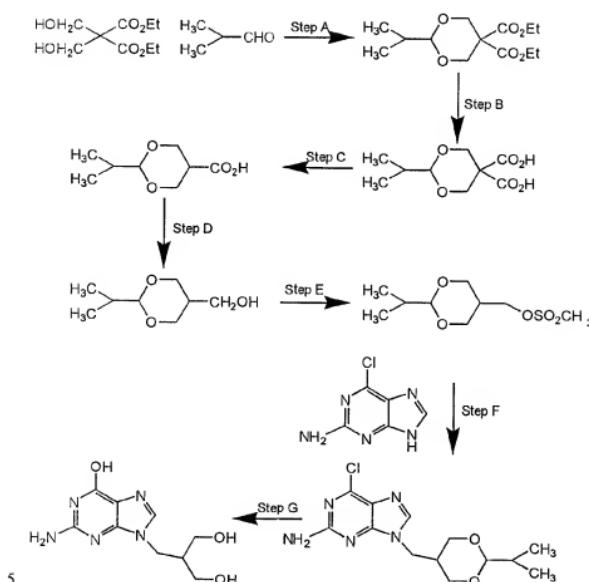
9-[3-Hydroxy-2-hydroxymethylprop-1-yl]-guanine

9-[3-Acetoxy-2-acetoxymethylprop-1-yl]-2-amino-6-iodopurine (2.5 g, 0.00576 mol) in 1.5M hydrochloric acid was heated at reflux for 3 hr. The reaction 20 mixture was cooled to room temperature and the pH adjusted to 14 with sodium hydroxide. The reaction mixture was stirred for a further 1.5 hr, then neutralized with concentrated hydrochloric acid. The resulting precipitate was filtered off and recrystallised from water to afford 9-[3-hydroxy-2-hydroxymethylprop-1-yl]-guanine (1.267 g, 92%) as colourless fluffy crystals. M.p. 294 - 296°. ¹H n.m.r. 25 (DMSO-d₆) 1.9 - 2.05, m, 1H; 3.3, t, *J* = 5.5 Hz, 4H; 3.95, d, *J* = 7.0 Hz, 2H; 4.6, t, *J* = 5.5 Hz, 2H; 6.5, s, 2H; 7.6, s, 1H; 10.1, s, 1H.

- 21 -

Example 1 (Alternate Route)Scheme 2

Diagrammatic representation of the alternate reaction sequence.



- 22 -

Step A

5,5-Dicarboxyethyl-2-isopropyl-1,3-dioxane

5

5,5-Dicarboxyethyl-2-isopropyl-1,3-dioxane was prepared according to a modified method of Eliel *et al.* A solution of diethyl bis(hydroxymethyl)malonate (25.0 g, 0.113 mol), isobutylaldehyde (0.226 mol, 20.6 ml) and p-toluenesulfonic acid (0.300 g) in petroleum spirit (50 ml) was heated at reflux and water collected in a Dean-Stark 10 apparatus. The petroleum spirit was removed and the product distilled to afford 5,5-dicarboxyethyl-2-isopropyl-1,3-dioxane (22.5 g, 73%) as a colourless oil. B.p. 103 /0.15 mm Hg. ^1H n.m.r. (CDCl_3) 0.9, d, $J = 6.9$ Hz, 6H; 1.2, t, $J = 6.9$ Hz, 3H; 1.3, t, $J = 6.9$ Hz, 3H; 1.7 - 1.85, m, 1H; 3.9, d, $JAB = 11.5$ Hz, 2H; 4.15, q, $J = 6.9$ Hz, 2H; 4.22, d, $J = 4.8$ Hz, ^1H ; 4.3, q, $J = 6.9$ Hz, 2H; 4.7, d, $JAB = 11.5$ Hz, 2H.

15

Step B

5,5-Dicarboxylic-2-isopropyl-1,3-dioxane

20 5,5-Dicarboxylic-2-isopropyl-1,3-dioxane was synthesised from 5,5-dicarboxyethyl-2-isopropyl-1,3-dioxane (20.0 g, 72.9 mmol) according to the method of Eliel *et al.* The crude product was re-crystallised from ethyl acetate and petroleum spirit to give 5,5-dicarboxylic-2-isopropyl-1,3-dioxane (14.1 g, 80%) as a colourless solid. M.p. 143.5 - 145°. ^1H n.m.r. (DMSO-d_6) 0.8, d, $J = 6.9$ Hz, 6H; 1.55 - 1.75, m, 1H; 3.8, d, $JAB = 11.3$ Hz, 2H; 4.3, d, $J = 4.8$ Hz, 1H; 4.5, d, $JAB = 11.3$ Hz, 2H; 12.8, br s, 2H.

Step C

5-Carboxy-2-isopropyl-1,3-dioxane

5-Carboxy-2-isopropyl-1,3-dioxane was synthesised from 5,5-dicarboxylic-2-isopropyl-1,3-dioxane (15.7 g, 64.8 mmol) according to the method of Eliel *et al.* The crude product was re-crystallised from ethyl acetate and petroleum spirit to give 5-carboxy-2-isopropyl-1,3-dioxane (9.75 g, 76%) as a colourless solid. M.p. 131-134°. ¹H n.m.r. (DMSO-d₆) 0.8, d, *J* = 6.9 Hz, 6H; 1.6 - 1.8, m, 1H; 2.7 - 2.85, m, 1H; 10 3.65, t, *JAB* = 11.5 Hz, 2H; 4.15, t, *JAB* = 10.1 Hz, 2H; 4.2, t, *J* = 4.8 Hz, 1H.

Step D

5-Hydroxymethyl-2-isopropyl-1,3-dioxane

15

To a solution of 5-carboxy-2-isopropyl-1,3-dioxane (8.75 g, 44.1 mmol) in anhydrous diethyl ether (65 ml) under a nitrogen atmosphere, was added borane methylsulfide complex (9.0 ml, 10M, 90 mmol). The reaction mixture was heated to reflux for 1 hr, cooled to room temperature and quenched with water (20 ml) and 20 methanol (30 ml). The methanol was removed and the aqueous phase extracted several times with diethyl ether. The combined diethyl ether extracts were washed with water and brine, dried over magnesium sulfate and the solvent removed to give 5-hydroxymethyl-2-isopropyl-1,3-dioxane (6.50 g, 80%) as a colourless oil. ¹H n.m.r. (DMSO-d₆) 0.85, d, *J* = 7 Hz, 6H; 1.6 - 1.8, m, 1H; 1.85 - 2.05, m, 1H; 3.2, t, *JAB* = 11.3 Hz, 2H; 3.3 25 - 3.45, m, 4H; 4.15, d, *J* = 4.8 Hz, 1H.

- 24 -

Step E

2-Isopropyl-5-(methanesulfoxy)methyl-1,3-dioxane

5 2-Isopropyl-5-(methanesulfoxy)methyl-1,3-dioxane was synthesised from 5-carboxy-2-isopropyl-1,3-dioxane (7.30 g, 39.6 mmol) according to the method used for 3-acetoxy-2-acetoxymethylprop-1-yl methanesulfonate to give the product as a colourless oil (10.1 g, 97%). ¹H n.m.r. (DMSO-d₆) 0.85, d, *J* = 6.9 Hz, 6H; 1.6 - 1.8, m, 1H; 2.15 - 2.35, m, 1H; 3.2, s, 3H; 3.5, t, *JAB* = 11.3 Hz, 2H; 4.0 - 4.15, m, 4H; 4.2, d, *J*₁₀ = 4.8 Hz, 1H.

Step F

2-Amino-6-chloro-9-[(2-isopropyl-1,3-dioxan-5-yl)methyl]purine

15

2-Amino-6-chloro-9-[(2-isopropyl-1,3-dioxan-5-yl)methyl]purine was synthesised from 2-isopropyl-5-(methanesulfoxy)methyl-1,3-dioxane (10.1 g, 38.5 mmol) according to method used for 9-[3-acetoxy-2-acetoxymethylprop-1-yl]-2-amino-6-iodopurine to give the product as a colourless solid (4.80 g, 40%). M.p. 204 - 205°. ¹H n.m.r. (DMSO-d₆) 0.85, d, *J* = 7 Hz, 6H; 1.6 - 1.75, m, 1H; 2.4 - 2.55, m, 1H; 3.45, t, *J* = 11.3 Hz, 2H; 3.85 - 4.0, m, 4H; 4.2, d, *J* = 5 Hz, 1H; 6.9, s, 2H; 8.1, s, 1H. ¹³C n.m.r. (DMSO-d₆) 16.8, 31.9, 34.3, 41.4, 48.6, 104.5, 123.3, 143.1, 149.4, 154.2, 159.7. Mass spectrum: *m/z* 312 ((M+1)+, 100%), 340 ((M+29)+, 15), 314 ((M+3)+, 30), 313 ((M+2)+, 18), 276 (20). Accurate mass: found 312.1209 (M+1)+, 25 C₁₃H₁₉N₅O₂C1, required 312.1227.

- 25 -

Step G

9-[3-Hydroxy-2-hydroxymethylprop-1-yl]-guanine

5 9-[1-Hydroxy-2-hydroxymethylpropyl]guanine was synthesised from 2-amino-6-chloro-9-[(2-isopropyl-1,3-dioxan-5-yl)methyl]purine (1.28 g, 4.11 mmol) according to the method used for 9-[3-hydroxy-2-hydroxymethylprop-1-yl]-guanine (step E above) to afford the product as a colourless solid (0.60 g, 61%). M.p. 285° dec. ^1H n.m.r. (DMSO-d₆) 1.9 - 2.05, m, 1H; 3.3, t, J = 5.5 Hz, 4H; 3.95, d, JJ = 7.0 Hz, 2H; 10 4.6, t, J = 5.5 Hz, 2H; 6.5, s, 2H; 7.6, s, 1H; 10.1, s, 1H. ^{13}C n.m.r.(DMSO-d₆) 41.5, 43.6, 58.9, 116.3, 138.1, 151.3, 153.4, 156.8. Mass spectrum: *m/z* 340 ((M+1)⁺, 100%), 368 ((M+29)⁺, 20), 341 ((M+2)⁺, 12).

Example 2

15

9-[3-Hydroxy-2-hydroxymethylprop-1-yl]-2-amino-6-methoxypurine

A mixture of 9-[3-acetoxy-2-acetoxymethylprop-1-yl]-2-amino-6-iodopurine (307 mg, 0.708 mmol), sodium hydroxide (7.75 g, 194 mmol), methanol (30 ml) and water

20 (3 ml) was stirred at room temperature for 18 hr. The reaction mixture was neutralised with aqueous HCl and the solvent was removed by rotary evaporation. Hot methanol was added to the residue and the solution decanted off and filtered through a short silica column. The filtrate was concentrated and recrystallised from water to afford 9-[3-hydroxy-2-hydroxymethylprop-1-yl]-2-amino-6-methoxypurine as colourless crystals 25 (179 mg, quantitative). ^1H nmr (DMSO-d₆) 1.93 - 2.16, m, 1H; 3.22 - 3.40, m, 4H; 3.95, s, 3H; 4.19, d, J = 7Hz, 2H; 4.69, t, J = 5Hz, 2H; 6.45, s, 2H; 7.79, s, 1H.

- 26 -

Example 3

9-[3-Acetoxy-2-acetoxymethylprop-1-yl]-2-amino-6-hydrazino-purine

5

A mixture of 9-[3-acetoxy-2-acetoxymethylprop-1-yl]-2-amino-6-iodopurine (300 mg, 0.69 mmol), hydrazine hydrate (85%, 210 μ l, 6.68 mmol) and ethanol (35 ml) under a nitrogen atmosphere, was stirred at reflux for 3 hr and then at room temperature for 16 hr. The resulting colourless solid was filtered off, washing well with ethanol and dried in 10 vacuo to afford 9-[3-acetoxy-2-acetoxymethylprop-1-yl]-2-amino-6-hydrazino-purine (196 mg, 84%). 1 H n.m.r. (DMSO-d₆) 1.98, s, 6H; 2.54 - 2.77, m, 1H; 3.87 - 4.08, m, 4H; 4.05, d, *J* = 7Hz, 2H; 4.40, br s, 2H; 5.89, br s, 2H; 7.67, s, 1H; 8.40, s, 1H.

Example 4

15

2-Amino-6-hydrazino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]purine

A mixture of 9-[3-acetoxy-2-acetoxymethylprop-1-yl]-2-amino-6-iodopurine (300 mg, 0.69 mmol), hydrazine hydrate (85%, 700 μ l, 22.27 mmol) and ethanol (35 ml) 20 under a nitrogen atmosphere, was stirred at reflux for 6 hr and then at room temperature for 16 hr. The reaction mixture was concentrated to dryness and the residue recrystallised from water to afford 2-amino-6-hydrazino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]purine (86 mg, 49%) as cream crystals. 1 H n.m.r. (DMSO-d₆) 1.90 - 2.12, m, 1H; 3.20 - 3.40, m, 4H; 3.98, d, *J* = 7Hz, 2H; 4.42, br s, 2H; 4.74, t, *J* = 5Hz, 2H; 5.98, 25 s, 2H; 7.62, s, 1H; 8.45, s, 1H.

- 27 -

Example 5

2-Amino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]-6-iodopurine

5

A mixture of 9-[3-acetoxy-2-acetoxymethylprop-1-yl]-2-amino-6-iodopurine (304 mg, 0.70 mmol) and methanolic ammonia (10 ml) were stirred for 2 hr in a stoppered flask. The stopper was removed and the reaction mixture left to stand for 16 hr. The reaction mixture was filtered giving 2-amino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]-6-10 iodopurine (201 mg, 82%) as colourless needles. ^1H n.m.r. (DMSO- d_6) 1.96 - 2.19, m, 1H; 3.25 - 3.43, m, 4H; 4.01, d, J = 7Hz, 2H; 4.61, t, J = 5Hz, 2H; 6.84, s, 2H; 8.02, s, 1H.

15

Example 6

2,6-Diamino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]purine

20 A mixture of 9-[3-acetoxy-2-acetoxymethylprop-1-yl]-2-amino-6-iodopurine (299 mg, 0.69 mmol) and methanolic ammonia (10 ml) were stirred for 18 hr in a bomb at 100°. The reaction mixture was cooled to room temperature and after 1 hr a precipitate formed. This was filtered off and dried in vacuo to afford 2,6-diamino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]purine (128 mg, 78%) as colourless crystals. ^1H n.m.r. 25 (DMSO- d_6) 1.90 - 2.10, m, 1H; 3.20 - 3.40, m, 4H; 3.96, d, J = 6.5Hz, 4.75, t, J =

- 28 -

5Hz, 2H; 5.84, s, 2H; 6.70, s, 2H; 7.78, s, 1H.

Example 7

5 2-(Dimethylaminomethylene)amino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]guanine

A mixture of 9-[3-hydroxy-2-hydroxymethylprop-1-yl]-guanine (200 mg, 0.84 mmol), N,N-dimethylformamide dimethyl acetal (1.5 ml, 11.3 mmol) and N,N-dimethylformamide (20 ml) under a nitrogen atmosphere was stirred at room temperature 10 for 2 days. Solvents were removed in vacuo at 60° and the residue was recrystallised from ethanol to afford 2-(dimethylaminomethylene)amino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]guanine (180 mg, 73%) as colourless crystals. ¹H n.m.r. (DMSO-d₆) 1.95 - 2.17, m, 1H; 3.03, s, 3H; 3.15, s, 3H; 3.21 - 3.47, m, 4H; 4.03, d, J = 7Hz, 2H; 4.62, t, J = 5Hz, 2H; 7.74, s, 1H; 8.53, s, 1H; 11.26, br s, 1H.

15

Example 8

2-Amino-9-[(2-isopropyl-1,3-dioxan-5-yl)methyl]purine

20 A mixture of 2-amino-6-chloro-9-[(2-isopropyl-1,3-dioxan-5-yl)methyl]purine (350 mg, 1.12 mmol), triethylamine (172 μ l, 1.23 mmol), 10% palladium on carbon (35 mg) and ethanol (5 ml) was stirred under an atmosphere of hydrogen for 3 days. The reaction mixture was then filtered through GFA paper washing copiously with dichloromethane. The filtrate was concentrated to dryness, dissolved in dichloromethane, 25 washed with water (2 x) and brine, dried over sodium sulfate and concentrated to afford

- 29 -

2-amino-9-[(2-isopropyl-1,3-dioxan-5-yl)methyl]purine (260 mg, 84%) as a colourless solid. ^1H n.m.r. (DMSO-d₆) 0.83, d, J = 7Hz, 6H; 2.35 - 2.63, m, 1H; 3.45, t, J = 11Hz, 2H; 3.87, d, J = 7Hz, 2H; 3.82 - 3.95, m, 2H; 4.17, d, J = 5Hz, 1H; 6.53, s, 2H; 8.02, s, 1H; 8.57, s, 1H.

5

Example 9

2-Amino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]purine

10 2-Amino-9-[(2-isopropyl-1,3-dioxan-5-yl)methyl]purine (160 mg, 0.58 mmol) was stirred in trifluoroacetic acid (5 ml) for 2 hr. After this time a few drops of water was added and the reaction mixture was stirred for a further 3 hr. Solvent was removed in vacuo and the residue was neutralized with saturated sodium bicarbonate. The solution was concentrated to dryness and hot methanol was added. The solution was decanted and 15 passed through a short silica column. The crude material was then hplc chromatographed with 2% acetonitrile in water as the eluting solvent . The fractions containing the desired product were combined and freeze dried to afford 2-amino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]purine (70 mg, 55%) as a fluffy colourless solid. ^1H n.m.r. (DMSO-d₆) 2.00 - 2.20, m, 1H, 3.20 - 3.40, m, 4H; 4.06, d, J = 7Hz, 2H; 4.68, br s, 20 2H; 6.53, s, 2H; 8.00, s, 1H; 8.57, s, 1H.

25

- 30 -

Example 10

9-[3-Hydroxy-2-hydroxymethylprop-1-yl]-guanine sodium salt

5 9-[3-Hydroxy-2-hydroxymethylprop-1-yl]-guanine (147.5 mg, 0.616 mmol) was dissolved in aqueous sodium hydroxide (1.0 M, 616 μ l, 0.616 mmol). The solution was filtered, washing with a small amount of water, then freeze dried to afford 9-[3-hydroxy-2-hydroxymethylprop-1-yl]-guanine sodium salt (160.7 mg, quantitative) as a fluffy colourless solid. 1 H n.m.r. (DMSO-d₆) 1.78 - 2.02, m, 1H; 3.06 - 3.29, m, 4H; 3.91, 10 d, J = 6Hz, 2H; 5.16, br s, 2H; 5.42, br s, 2H; 7.32, s, 1H.

Example 11

9-[3-Acetoxy-2-hydroxymethylprop-1-yl]-guanine

15 To a solution of 9-[3-hydroxy-2-hydroxymethylprop-1-yl]-guanine (1.0 g, 4.18 mmol) in N,N-dimethylformamide (5 ml) under a nitrogen atmosphere, was added trifluoroacetic acid (510 μ l, 6.63 mmol) and triethylorthoacetate (790 μ l, 4.31 mmol) resulting in a cloudy mixture. The reaction was monitored by hplc (5% acetonitrile in 20 water) and more triethylorthoacetate (720 μ l, 3.92 mmol) was added portionwise until the reaction was complete. Water (243 μ l, 13.5 mmol) was added and the reaction stirred for a further 1.5 hr. The reaction mixture was neutralised with sodium bicarbonate and solvent was removed at room temperature. The crude product was recrystallised from water to give a colourless product (901 mg). This was preadsorbed and flash 25 chromatographed on silica eluting with 10%, 15% and 17 % methanol in dichloromethane. The fractions containing pure product were combined and concentrated

- 31 -

to afford 9-[3-acetoxy-2-hydroxymethylprop-1-yl]-guanine (520 mg, 44 %) as a colourless solid. ^1H n.m.r. (DMSO-d₆) 1.94, s, 3H; 2.20 - 2.40, m, 1H; 3.26 - 3.43, m, 2H; 3.84 - 4.03, m, 4H; 4.82, t, J = 5Hz, 1H; 6.46, s, 2H; 7.64, s, 1H; 10.58, s, 1H.

5

Example 12

9-[3-Hydroxy-2-hydroxymethylprop-1-yl]-guanine triphosphate tetraammonium salt

10 The procedure used for the preparation followed that of Ludwig *et al.* A solution of 2-chloro-4H-1,3,2-benzodioxaphosphorin-4-one (150 mg, 0.74 mmol) in dry dioxane (2 ml) was added dropwise to the stirred 9-[3-acetoxy-2-hydroxymethylprop-1-yl]-guanine (187.8 mg, 0.668 mmol, dried under high vac. at 85°C for *ca.* 7-8 h) in dry N,N-dimethylformamide (10 ml) and dry pyridine (2 ml) over approximately 5 min. Stirring 15 was continued for 0.75 hr. Bis[(tri-*n*-butyl)ammonium]pyrophosphate hemi DMF (590 mg; 1.0 mmol) dissolved in dry N,N-dimethylformamide (2.5 ml) containing dry *n*-Bu₃N(0.75 ml) was then added dropwise to the stirred solution.

After *ca.* 2.5 h stirring at room temperature, the yellow reaction solution was treated 20 dropwise with iodine solution (9.5 ml) made by dissolving iodine (3.56 g) in pyridine (200 ml) and water (5 ml). The slight excess of iodine added was destroyed on addition of a few drops of 5% NaHSO₃ solution.

After stirring for *ca.* 1.5 hr at room temperature, the solvent was removed below 30° to 25 give an orange-yellowish oil which was treated with 25 ml water at room temperature for 1 h with strong stirring. Concentrated NH₄OH (50 ml) was added and the reaction

- 32 -

mixture was stirred for 3 hr at room temperature before removing the NH₃OH solution at 25°C. The semi-solid product was treated twice with acetone to give a pale yellow solid (0.56 g).

The product (0.5 g) was dissolved in a little water, centrifuged and made up to 5 ml in 5 volume and HPLC chromatographed in lots of 0.5 ml eluting with water (flow rate of 12 ml/min). The fractions between 13-14 mins and 18-19 mins were collected and were freeze dried to give a pale yellow solid (320 mg).

The material was dissolved in water (4 ml) and re-chromatographed in 0.5 ml lots. For each fraction the leading and trailing section of the peak containing the triphosphate was 10 cut. This was repeated a further 4 times. Finally the product was dialysed (100 MWCO tubing, 6.5 h, H₂O) before again purifying by HPLC. This treatment removed a small amount of phosphorus impurity (small ³¹P nmr peak at δ +0.93), due, presumably, to some inorganic phosphate.

15 ¹H nmr (D₂O); δ 2.41, s, 1H; 3.60, d, *J*=5.38 Hz, 2H; overlapping doublets 4.00, d, *J*=4.98 Hz and 4.17, d, *J*=5.76 Hz, 4H; 7.87, s, 0.8H.

³¹P nmr (D₂O): In the nmr of the crude triphosphate the phosphate peaks were the best defined; *viz.* P_β (t, δ-21.16, *J_{pp}*=19.66 Hz); P_α (doublet of triplets at δ-9.87 (*J_{pp}* 19.40 Hz; *J_{PH}* 5.39 Hz); P_α (d, δ-5.96; *J_{pp}* 20.01 Hz). The ³¹P nmr peaks broadened and moved on HPLC purification so that the purified sample had two broad ³¹P peaks at δ-9.96 (P_α+P_α) and δ-22.07 (P_β) of intensity ratio 2:1.

- 33 -

Example 13

2-Amino-6-cyclopropylamino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]purine

5 A mixture of 9-[3-acetyloxy-2-acetyloxymethylprop-1-yl]-2-amino-6-iodopurine (557 mg, 1.24 mmol) and cyclopropylamine (1.0 ml, 14.4 mmol) were stirred for 18 hr in a bomb at 80°. The reaction mixture was cooled to room temperature and concentrated to give a crude orange oil (989 mg). A portion of this was then hplc chromatographed with 3% acetonitrile in water as the eluting solvent. The fractions containing the desired product were combined 10 and freeze dried to afford 2-amino-6-cyclopropylamino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]purine as yellow oil which solidified on standing. ¹H n.m.r. (DMSO-d₆) 0.55 - 0.76, m, 4H; 1.91 - 2.12, m, 1H; 2.95 - 3.13, m, 1H; 3.21 - 3.45, m, 4H; 3.97, d, *J* = 7 Hz, 2H; 4.75, t, *J* = 5 Hz, 2H; 5.91, br s, 2H; 7.09, d, *J* = 5 Hz, 1H; 7.62, s, 1H.

15 Example 14

9-[3-Acetyloxy-2-acetyloxymethylprop-1-yl]-2-aminopurine

9-[3-Acetyloxy-2-acetyloxymethylprop-1-yl]-2-aminopurine was prepared according to the 20 method of Example 8 from 9-[3-acetyloxy-2-acetyloxymethylprop-1-yl]-2-amino-6- iodopurine (5.0 g, 11.5 mmol), triethylamine (1.76 ml, 12.65 mmol) and 10% palladium on carbon (500 mg) in ethanol (200 ml). Yield: 2.33 g (66%). ¹H n.m.r. (DMSO-d₆) 1.93, s, 6H; 2.59 - 2.80, m, 1H; 3.96, dd, *J* = 5.6 & 11.4 Hz, 2H; 4.03, dd, *J* = 5.6 & 11.4 Hz, 2H; 4.15, d, *J* = 7 Hz, 2H; 6.49, br s, 2H; 8.04, s, 1H; 8.57, s, 1H.

Example 15

9-[3-Acetyloxy-2-acetyloxymethylprop-1-yl]-guanine

5 A mixture of 9-[3-hydroxy-2-hydroxymethylprop-1-yl]-guanine (560 mg, 2.34 mmol) and 4-dimethylaminopyridine (50 mg) in acetic anhydride (15 ml) was stirred at room temperature for 16 hr. The reaction mixture was concentrated to dryness and the residue was partitioned between water and chloroform. The resulting solid was filtered off and recrystallised from methanol to afford 9-[3-acetyloxy-2-acetyloxymethylprop-1-yl]-guanine as colourless 10 crystals (593 mg, 78%). ^1H n.m.r. (DMSO- d_6) 1.97, s, 6H; 2.53 - 2.70, m, 1H; 3.87 - 4.09, m, 6H; 6.42, br s, 2H; 7.67, s, 1H; 10.56, br s, 1H.

Example 16 & 17

15 9-[2-L-Valyloxymethyl-3-L-valyloxyprop-1-yl]-guanine bishydrochloride salt and 9-[3-Hydroxy-2-L-valyloxymethylprop-1-yl]-guanine hydrochloride salt

20 A mixture of 9-[3-hydroxy-2-hydroxymethylprop-1-yl]-guanine (3.02 g, 12.6 mmol), N-benzyloxycarbonyl-L-valyl-N-carboxy anhydride (Z-L-valyl-NCA) (purchased from Isochem 20 or SNPE North American Inc.) (3.69 g, 13.32 mmol) and 4-dimethylaminopyridine (75 mg) in N,N-dimethylformamide (75 ml), under a nitrogen atmosphere, was stirred at room 24 temperature for 16 hr. HPLC analysis indicated incomplete reaction. A further portion of Z-L-valyl-NCA (1.84 g, 6.67 mmol) was added and the reaction mixture was stirred a further 24 hr. Concentrated to dryness and ethyl acetate was added to precipitate out unreacted 25 9-[3-hydroxy-2-hydroxymethylprop-1-yl]-guanine. The filtrate was concentrated, preadsorbed onto silica and flash chromatographed on silica with 5%, 7%, 10% & 15% methanol in dichloromethane as the eluting solvents. Fractions containing a single spot on tlc were combined to give 9-[2-(N-benzyloxycarbonyl-L-valyloxymethyl)-3-(N-benzyloxycarbonyl-L-valyloxyprop-1-yl] guanine as a colourless solid (2.54 g). ^1H n.m.r. (DMSO- d_6) 0.86, d,

- 35 -

12H; 1.85 - 2.15, m, 2H; 2.51 - 2.70, m, 1H; 3.80 - 4.18, m, 8H; 5.04, s, 4H; 6.41, br s, 2H; 7.31, br s, 10H; 7.59, s, 1H; 7.75, d, J = 8 Hz, 2H; 10.62, s, 1H.

The remaining fractions were combined and rechromatographed with 5%, 7%, 10% & 15% 5 methanol in dichloromethane as the eluting solvents to give a further 229 mg of the divaly 1 compound and a colourless solid (880 mg). This was dissolved in dichloromethane then washed with saturated sodium bicarbonate (2x) and brine, then dried over magnesium sulfate to give 9-[2-(N-benzyloxycarbonyl-L-valyloxy methyl)-3-hydroxyprop-1-yl] guanine as a colourless foamy solid (266 mg) 1 H n.m.r. (DMSO-d₆) 0.86, d, J = 6.8 Hz, 6H; 1.88 - 10 2.11, m, 1H; 2.20 - 2.39, m, 1H; 3.21 - 3.43, m, 2H; 3.78 - 4.10, m, 5H; 4.82, t, J = 5 Hz, 1H; 5.04, s, 2H; 6.46, br s, 2H; 7.34, br s, 5H; 7.61, d, J = 3 Hz, 1H; 7.65 - 7.75, m, 1H; 10.59, br s, 1H.

9-[2-L-Valyloxymethyl-3-L-valyloxyprop-1-yl] guanine bishydrochloride salt

15

A mixture of 9-[2-(N-benzyloxycarbonyl-L-valyloxy methyl)-3-(N-benzyloxycarbonyl-L-valyloxyprop-1-yl]guanine (858 mg, 1.22 mmol), 1M aqueous hydrochloric acid (2.44 ml, 2.43 mmol) and 10% palladium on carbon (215 mg) in ethanol (50 ml) was stirred in an atmosphere of hydrogen for 3 hr. The reaction mixture was filtered through GFA paper 20 washing copiously with ethanol. The filtrate was concentrated to dryness at 40°. The residue was taken up in water and filtered through a short column packed with GFA paper, Celite 577 and more GFA paper. The filtrate was freeze dried to give 9-[2-L-valyloxymethyl-3-L-valyloxyprop-1-yl] guanine bishydrochloride salt as a cream solid (517 mg, 83%). 1 H n.m.r. (DMSO-d₆) 0.87 - 1.02, m, 12H; 2.05 - 2.30, m, 2H; 2.60 - 2.80, m, 1H; 3.85, dd, J = 4.6 25 & 11.1 Hz, 2H; 4.00 - 4.32, m, 6H; 6.59, s, 2H; 7.78, s, 1H; 8.63, br s, 6H; 10.78, br s, 1H.

- 36 -

9-[3-Hydroxy-2-L-valyloxymethylprop-1-yl]guanine hydrochloride salt

9-[3-Hydroxy-2-L-valyloxymethylprop-1-yl] guanine hydrochloride salt was prepared according to the method of Example 16 from 9-[2-(N-benzyloxycarbonyl-L-valyloxy 5 methyl)-3-hydroxyprop-1-yl]-guanine (212 mg, 0.45 mmol), 1M aqueous hydrochloric acid (0.45 ml, 0.45 mmol) and 10% palladium on carbon (56 mg) in ethanol (15 ml) to give the product as a colourless fluffy solid (162 mg, 96%). ¹H n.m.r. (DMSO-d₆) 0.96, dd, *J* = 7.2 & 11.9 Hz, 6H; 2.00 - 2.25, m, 1H; 2.25 - 2.47, m, 1H; 3.31 - 3.51, m, 2H; 3.82, dd, *J* = 4.6 & 13.5 Hz, 1H; 3.96 - 4.14, m, 4H; 4.91, br s, 1H; 6.59, br s, 2H; 7.69, s, 1H, 8.52, br s, 10 3H; 10.72, br s, 1H.

Example 18

9-[3-Acetoxy-2-L-valyloxymethyl]prop-1-yl]-guanine hydrochloride salt

15

A mixture of 9-[3-acetoxyl-2-hydroxymethylprop-1-yl] guanine (330 mg, 1.17 mmol), Z-L-valyl-NCA (358 mg, 1.29 mmol) and 4-dimethylaminopyridine (30 mg) in N,N-dimethylformamide (20 ml), under a nitrogen atmosphere, was stirred at room temperature for 2 days. The reaction mixture was concentrated to dryness and the residue preadsorbed 20 onto silica and flash chromatographed with 10% methanol in dichloromethane. Fractions containing the desired product were combined and concentrated to give 9-[3-acetoxyl-2-(N-benzyloxycarbonyl-L-valyloxy methyl)prop-1-yl] guanine as a colourless solid (313 mg, 52%). ¹H n.m.r. (DMSO-d₆) 0.87, d, *J* = 7 Hz, 6H; 1.97, s, 3H; 1.88 - 2.13, m, 1H; 2.53 - 2.71, m, 1H; 3.82 - 4.15, m, 7H; 5.04, s, 2H; 6.43, br s, 2H; 7.35, br s, 5H; 7.63, d, *J* = 3.6 25 Hz, 1H; 7.75, d, *J* = 8.1 Hz, 1H.

9-[3-Acetoxy-2-L-valyloxymethyl]prop-1-yl]-guanine hydrochloride salt was prepared according to the method of Example 16 from 9-[3-acetoxyl-2-(N-benzyloxycarbonyl-L-

- 37 -

valyloxymethyl)prop-1-yl]-guanine (201 mg, 0.39 mmol), 1M aqueous hydrochloric acid (0.39 ml, 0.39 mmol) and 10% palladium on carbon (62 mg) in ethanol (10 ml) to give the product as a colourless fluffy solid (162 mg, quantitative). ¹H n.m.r. (DMSO-d₆) 0.95, dd, J = 3.8 & 6.8 Hz, 6H; 1.99, s, 3H; 2.03 - 2.25, m, 1H; 2.57 - 2.78, m, 1H; 3.84, dd, J = 4.7

5 & 10.2 Hz, 1H; 3.93 - 4.23, m, 6H; 6.54, br s, 2H; 7.71, s, 1H; 8.33, br s, 3H; 10.71, br s, 1H.

Example 19

10 9-[3-Hydroxy-2-palmityloxymethylprop-1-yl] guanine

Palmitoyl chloride (2.07 g, 7.53 mmol) was dissolved in dry dichloromethane and made up to a volume of 10 ml and used as a stock solution.

To a suspension of 9-[3-hydroxy-2-hydroxymethylprop-1-yl] guanine (1.0 g, 4.18 mmol) in 15 pyridine (20 ml) and N,N-dimethylformamide (10ml), under a nitrogen atmosphere, was added the stock solution of palmitoyl chloride (3.5 ml). The reaction mixture was stirred for 16 hr at room temperature and a further aliquot (3.5 ml) of stock solution was added. The reaction mixture was stirred for 24 hr and the final aliquot of stock solution was added. The reaction mixture was stirred for 2 more days, the solvents were removed *in vacuo* at 60°.

20 The crude solid (3.7 g) was preadsorbed and flash chromatographed on silica eluting with 5% to 23% methanol in dichloromethane. The fractions containing pure product were combined to give 9-[3-hydroxy-2-palmityloxymethylprop-1-yl] guanine as a colourless solid (595 mg). ¹H n.m.r. (DMSO-d₆) 0.85, t, J = 6.8 Hz, 3H; 1.12 - 1.38, m, 24H; 1.33 - 1.60, m, 2H; 2.19, q, J = 7.2 Hz, 2H; 3.32 - 3.40, m, 2H; 3.95, t, J = 6 Hz, 2H; 4.07 - 4.20, 25 m, 2H; 4.87, br s, 1H; 6.65, br s, 2H; 7.63, s, 1H; 10.76, br s, 1H.

Example 20

9-[3-Palmityloxy-2-L-valyloxymethylprop-1-yl] guanine hydrochloride salt

A mixture of 9-[3-hydroxy-2-palmityloxyethylprop-1-yl] guanine (534 mg, 1.12 mmol), Z-L-valyl-NCA (620 mg, 2.24 mmol) and 4-dimethylaminopyridine (25 mg) in N,N-

5 dimethylformamide (25 ml), under a nitrogen atmosphere, was stirred at room temperature for 16 hr. Solvent was removed *in vacuo* at 60° and the residue partitioned between dichloromethane and water. The layers were separated and the aqueous was extracted twice more with dichloromethane. The combined organic layers were washed with saturated sodium bicarbonate solution (2x) and brine, dried over magnesium sulfate and concentrated 10 to give the crude product as a colourless oil (509 mg). This was flash chromatographed on silica eluting with 5% methanol in dichloromethane. The fractions containing the pure product were combined to give 9-[3-palmityloxy-2-(N-benzyloxycarbonyl-L-valyloxymethyl)prop-1-yl] guanine as a colourless foamy solid (290 mg). ¹H n.m.r. (DMSO-d₆) 0.75-0.95, m, 9H; 1.22, br s, 24H; 1.33 - 1.60, m, 2H; 1.90 - 2.15, m, 1H; 15 2.23, t, *J* = 7.2 Hz, 2H; 2.50 - 2.75, m, 1H; 3.85 - 4.14, m, 7H; 5.04, s, 2H; 6.45, br s, 5H; 7.62, d, *J* = 3.8 Hz, 1H; 7.74, d, *J* = 8.2 Hz, 1H; 10.66, br s, 1H.

9-[3-Palmityloxy-2-L-valyloxymethylprop-1-yl] guanine hydrochloride salt was prepared according to the method of Example 16 from 9-[3-palmityloxy-2-(N-benzyloxycarbonyl-L-valyloxymethyl)prop-1-yl] guanine (174 mg, 0.24 mmol), 1M aqueous hydrochloric acid (0.24 ml, 0.24 mmol) and 10% palladium on carbon (50 mg) in ethanol (10 ml) to give the product as a colourless fluffy solid (124 mg, 84%). ¹H n.m.r. (DMSO-d₆) 0.78 - 0.98, m, 9H; 1.22, br s, 24H; 1.37 - 1.58, m, 2H; 1.99 - 2.22, m, 1H; 2.25, t, *J* = 7.2 Hz, 2H; 2.55 - 2.76, m, 1H; 3.74, dd, *J* = 4.8 & 10.0 Hz, 1H; 3.90 - 4.20, m, 6H; 6.51, br s, 2H; 7.30 - 25 8.10, br s, 3H; 7.69, s, 1H; 10.88, br s, 1H.

Example 21

9-[3-Hydroxy-2-cholyloxymethylprop-1-yl]-guanine

5 To a solution of cholic acid (1.71 g, 4.18 mmol) and diisopropylethylamine (567 mg, 4.39 mmol) in N,N-dimethylformamide (25 ml), under a nitrogen atmosphere, at 10°, was added ethyl chloroformate (400 μ l, 4.18 mmol) This was stirred for 30 minutes, then a mixture of 9-[3-hydroxy-2-hydroxymethylprop-1-yl] guanine (1.0 g, 4.18 mmol) in N,N-dimethylformamide (100 ml) was added and the reaction mixture was stirred for a further 2 days. HPLC analysis (70% methanol in water) indicated ~ 20 - 30% product had formed. The reaction mixture was concentrated to dryness and methanol was added to precipitate out unreacted 9-[3-hydroxy-2-hydroxymethylprop-1-yl] guanine. The filtrate was concentrated to dryness to give a yellow oil (2.0 g), which was purified by semi-preparative HPLC eluting with 70% methanol in water. 9-[3-Hydroxy-2-cholyloxymethylprop-1-yl] guanine was obtained as a colourless glassy solid (124 mg). 1 H n.m.r. (DMSO-d₆) 0.57, s, 3H; 0.80, s, 3H; 0.70 - 2.40, m, 27H; 3.08 - 3.28, m, 2H; 3.37, d, *J* = 5.2 Hz, 2H; 3.61, s, 1H; 3.78, s, 1H; 3.87 - 4.05, m, 5H; 4.12, d, *J* = 3.4 Hz, 1H; 4.33, d, *J* = 4.3 Hz, 1H; 4.82, t, *J* = 4.5 Hz, 1H; 6.52, s, 2H; 7.63, s, 1H; 10.74, br s, 1H.

10

15

20 Example 22

9-[3-Cholyoxy-2-L-valyloxy-methylprop-1-yl] guanine hydrochloride salt

25 9-[2-(N-Benzylloxycarbonyl-L-valyloxy-3-cholyloxymethylprop-1-yl]-guanine was prepared according to the method of Example 20 from crude 9-[3-hydroxy-2-cholyloxymethylprop-1-yl]-guanine (1.21 g, 1.92 mmol), Z-L-valyl-NCA (0.99 g, 3.57 mmol) and 4-dimethylaminopyridine (20 mg) in N,N-dimethylformamide (10 ml). The crude material was flash chromatographed on silica eluting with 10% methanol in dichloromethane. The fractions containing the pure product were combined to give 9-[2-(N-benzylloxycarbonyl-L-

- 40 -

valyloxy-3-cholyloxyethylprop-1-yl] guanine as a colourless foamy solid (258 mg). ¹H n.m.r. (DMSO-d₆) 0.57, s, 3H; 0.80, s, 3H; 0.74 - 2.40, m, 34 H; 2.50 - 2.78, m, 1H; 3.05 - 3.30, m, 2H; 3.60, s, 1H; 3.77, s, 1H; 3.83 - 4.20, m, 8H; 4.33, d, *J* = 4.3 Hz, 1H; 5.04, s, 2H; 6.44, br s, 2H; 7.34, br s, 5H; 7.62, d, *J* = 3.7 Hz, 1H; 7.74, d, *J* = 8.2 Hz, 1H; 10.61, 5 br s, 1H.

9-[3-Cholyloxy-2-L-valyloxy-methylprop-1-yl] guanine hydrochloride salt was prepared according to the method of Example 16 from 9-[2-(N-benzyloxycarbonyl-L-valyloxy-3-cholyloxyethylprop-1-yl] guanine (248 mg, 0.29 mmol), 1M aqueous hydrochloric acid 10 (0.29 ml, 0.29 mmol) and 10% palladium on carbon (30 mg) in ethanol (10 ml) to give the product as a cream solid

(185 mg, 84%). ¹H n.m.r. (DMSO-d₆) 0.57, s, 3H; 0.80, s, 3H; 0.74 - 2.40, m, 34H; 2.50 - 2.78, m, 1H; 3.05 - 3.30, m, 2H; 3.60, s, 1H; 3.77, s, 1H; 3.85, dd, *J* = 4.6 & 10.5 Hz, 1H; 3.93 - 4.26, m, 7H; 4.34, d, *J* = 4.0 Hz, 1H; 6.56, br s, 2H; 7.71, s, 1H; 8.25 - 8.80, br s, 15 3H; 10.72, br s, 1H.

Example 23

9-[2-Elaidyloxy-3-hydroxy-methylprop-1-yl] guanine

20

9-[2-Elaidyloxy-3-hydroxy-methylprop-1-yl] guanine was prepared according to the method of Example 19 from 9-[3-hydroxy-2-hydroxymethylprop-1-yl] guanine (1.3 g, 5.43 mmol), elaidoyl chloride (3.1 g, 9.70 mmol) dissolved in dry dichloromethane (7 ml), in pyridine (25 ml) and N,N-dimethylformamide (12 ml) After work-up, some elaidic acid was removed by 25 distillation, to give a crude yellow solid (2.14 g). This was chromatographed on silica eluting with 7.5% methanol in dichloromethane. The fractions containing pure product were combined to give 9-[2-elaidyloxy-3-hydroxy-methylprop-1-yl] guanine (640 mg). ¹H n.m.r. (DMSO-d₆) 0.84, t, *J* = 6.7 Hz, 3H; 1.23, br s, 20H; 1.30 - 1.59, m, 2H; 1.85 - 2.00, m,

- 41 -

4H, 2.20, t, J = 7.3 Hz, 2H, 2.22 - 2.41, m, 1H; 3.36, d, J = 5.4 Hz, 2H; 3.85 - 4.07, m, 4H; 4.82, t, J = 5 Hz, 1H; 5.23 - 5.48, m, 2H; 6.44, br s, 2H; 7.63, s, 1H; 10.58, br s, 1H

Example 24

5

9-[2-Stearoyloxy-3-valyloxymethyl]prop-1-yl] guanine hydrochloride salt

A mixture of 9-[2-elaidyloxy-3-hydroxymethylprop-1-yl] guanine (200 mg, 0.40 mmol), Z-L-valyl-NCA (121 mg, 0.44 mmol) and 4-dimethylaminopyridine (5 mg) in dichloromethane 10 (20 ml), under a nitrogen atmosphere, was stirred at room temperature for 3 days. A further portion of Z-L-valyl-NCA (40 mg, 0.14 mmol) was added and the reaction was stirred at 40° for 6 hr and then at room temperature for 16 hr. Solvent was removed *in vacuo* at 60°, the residue preadsorbed and flash chromatographed on silica eluting with 6% methanol in dichloromethane. The fractions containing the pure product were combined to give 9-[2-(N-15 benzylloxycarbonyl-L-valyloxy-3-elaidyloxy-methyl)prop-1-yl]-guanine as a colourless foamy solid (174 mg, 60%). ^1H n.m.r. (DMSO-d₆) 0.78 - 0.93, m, 9H; 1.22, br s, 20H; 1.30 - 1.59, m, 2H; 1.89 - 2.13, m, 5H; 2.23, t, J = 7.2, Hz, 2H; 2.50 - 2.72, m, 1H; 3.85 - 4.14, m, 6H; 5.04, s, 2H; 5.24 - 5.45, m, 2H; 6.43, s, 2H; 7.34, br s, 5H; 7.61, d, J = 3.9 Hz, 1H; 7.74, d, J = 8.1 Hz, 1H; 10.64, br s, 1H.

20

9-[2-Stearoyloxy-3-valyloxymethyl]prop-1-yl] guanine hydrochloride salt was prepared according to the method of Example 16 from 9-[2-(N-benzylloxycarbonyl-L-valyloxy-3-elaidyloxy-methyl)prop-1-yl] guanine (162 mg, 0.22 mmol), 1M aqueous hydrochloric acid (0.22 ml, 0.22 mmol) and 10% palladium on carbon (60 mg) in ethanol (10 ml) to give the 25 product as a colourless fluffy solid (108 mg, 77%). ^1H n.m.r. (DMSO-d₆) 0.84, t, J = 6.3 Hz, 3H; 0.93, dd, J = 3.2 & 6.8 Hz, 6H, 1.22 br s, 28H; 1.36 - 1.57, m, 2H; 1.98 - 2.23, m, 1H; 2.25, t, J = 7.3 Hz, 2H; 2.55 - 2.75, m, 1H; 3.77, dd, J = 4.6 & 10.0 Hz, 1H; 3.90 - 4.22, m, 6H; 6.51, br s, 2H; 7.69, s, 1H; 7.65 - 8.15, br s, 3H; 10.48 - 10.82, br s, 1H.

Example 25

2-Amino-9-[2-L-valyloxy-3-L-valyloxymethylprop-1-yl]purine bishydrochloride salt

5 A mixture of 2-amino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]purine (9)(500 mg, 2.24 mmol), Z-L-valyl-NCA (1.30 g, 4.70 mmol) and 4-dimethylaminopyridine (5 mg) in N,N-dimethylformamide (10 ml), under a nitrogen atmosphere, was stirred at room temperature for 18 hr. The reaction mixture was concentrated to dryness and the residue chromatographed on silica with ethyl acetate as the eluting solvent. The fractions containing 10 pure product were combined and concentrated to give 2-amino-9-[2-(N-benzyloxycarbonyl-L-valyloxymethyl)-3-(N-benzyloxycarbonyl-L-valyloxyprop-1-yl]purine as a honey coloured glassy solid (1.12 g, 73%). ^1H n.m.r. (DMSO- d_6) 0.86, d, J = 6.7 Hz, 12H; 1.85 - 2.15, m, 2H; 2.57 - 2.78, m, 1H; 3.83 - 4.19, m, 8H; 5.04, s, 4H; 6.49, s, 2H; 7.34, br s, 10H; 7.76, d, J = 8.2 Hz, 2H; 7.96, s, 1H; 8.59, s, 1H.

15

2-Amino-9-[2-L-valyloxy-3-L-valyloxymethylprop-1-yl]purine bishydrochloride salt was prepared according to the method of Example 16 from 2-amino-9-[2-(N-benzyloxycarbonyl-L-valyloxymethyl)-3-(N-benzyloxycarbonyl-L-valyloxyprop-1-yl]purine (500 mg, 0.73 mmol), 1M aqueous hydrochloric acid (1.45 ml, 1.45 mmol) and 10% palladium on carbon 20 (90 mg) in ethanol (25 ml) to give the product as a colourless fluffy solid (319 mg, 89%). ^1H n.m.r. (DMSO- d_6) 0.87 - 1.03, m, 12H; 2.03 - 2.27, m, 2H; 2.70 - 2.80, m, 1H; 3.83, dd, J = 4.8 & 11.2 Hz, 2H; 4.07 - 4.42, m, 6H; 6.50, s, 2H; 8.15 - 8.88, br s, 3H; 8.20, s, 1H; 8.60, s, 1H.

25 Example 26

9-[3-Acetoxy-2-acetoxymethylprop-1-yl]-6-thioguanine

A mixture of 9-[3-acetoxy-2-acetoxymethylprop-1-yl]-2-amino-6-iodopurine (576 mg, 1.33

- 43 -

mmol) and thiourea (100 mg, 1.33 mmol) in ethanol (7 ml) under a nitrogen atmosphere, was heated at reflux for 1 hr. The reaction mixture was chilled and the product filtered off, washing with ethanol. The pale lemon solid was dried in vacuo at 80° to give 9-[3-acetoxy-2-acetoxymethylprop-1-yl]-6-thioguanine (288 mg, 64%). ¹H n.m.r. (DMSO-d₆) 1.98, s, 5 6H; 2.52 - 2.74, m, 1H; 3.87 - 4.13, m, 6H; 6.77, s, 2H; 7.88, s, 1H; 11.89, s, 1H.

Example 27 Antiviral Activity

Tests of antiviral activity in human cells infected with hepatitis B were performed 10 according to the method of Korba and Gerin. The effective concentration for 50% and 90% inhibition of the replication of the virus was determined from dose response curves. Results for some compounds of the invention are shown in Table 2.

Table 2

15

Test Compound	EC ₅₀ μ M	EC ₉₀ μ M
Example 1	0.16	1.9
Example 13	4.4	13

20 Example 28

Bioavailability Testing

The oral bioavailability of various compounds of the invention was compared in rats. 25 Briefly, the compounds were administered by oral gavage at 0.2mmol/kg of body weight. The compounds were suspended in 1 mL of an aqueous vehicle containing 1% carboxymethylcellulose and 0.05% Tween 80. Plasma was sampled over an 8 hour period and the concentration of the parent compound, in this case the compound of

example 1, was determined by hplc.

Aliquots of rat plasma (150 μ L) were acidified with 10% trichloroacetic acid (37.5 μ L) and centrifuged at 3000 rpm for 10 min. The supernatant was filtered through 0.22 μ m cellulose acetate centrifuge filters. Samples (100 μ L) were then injected onto the C18

5 Waters Symmetry HPLC column (3.9 and 150 mm, 5 μ m) equilibrated at 40°C. The two component mobile phase (A – 0.05% trifluoroacetic acid and 20 mM heptane sulfonic acid in distilled, deionised water; B – 0.05% trifluoroacetic acid, 20 mM heptane sulfonic acid and 70% acetonitrile in distilled, deionised water) was pumped at 0.5 mL/min with the percentage of mobile phase component B increasing linearly from 0 to 25% over 25 min

10 Analysis was by ultraviolet detection at 254 nm. The parent compound eluted at 17 min.

The concentration of drug versus time profile was plotted and the area under the curve determined. This was compared with the area under the curve provided by intravenous administration of the sodium salt of the parent compound to provided a measure of total bioavailability as a percentage. Results are shown in Table 3.

15

	Test Compound	% Bioavailability
20	Example 1	2.8
	Example 9	53.7
	Example 14	66.2
	Example 16	16.3
	Example 17	33.1
	Example 18	6.5
	Example 20	21

25

It will be appreciated by persons skilled in the art that numerous variations and/or modifications may be made to the invention as described without departing from the spirit 30 or the scope of the invention. The present examples and specific details are, therefore, to

- 45 -

be considered in all respects as illustrative of the invention and not restrictive.

The steps, features, compositions and compounds disclosed herein or referred to or indicated in the specification and/or claims of this application, individually, collectively, and any and all combinations of any two or more of said steps or features.

5 Throughout this specification and the claims which follow, unless the context requires otherwise, the word "comprise", and variations such as "comprises" and "comprising", will be understood to imply the inclusion of a stated integer or step or group of integers or steps but not the exclusion of any other integer or step or group of integers or steps.

10

References:

Cullen *et al.*, *Antimicrob. Agents Chemother.*, **41** (10), 2076-82 (1997)

Martin *et al.*, *J. Med. Chem.* **29**, 1384-1389 (1986).

Greene T. W., *Protective Groups in Organic Synthesis*, John Wiley and Sons, Inc. (1991)

15 Bisacchi *et al.*, *J. Org. Chem.* **60**, 2902 - 2905 (1995).

Harnden *et al.*, *J. Med. Chem.* **33**, 187-196 (1990)

Korba and Gerin, *Antiviral Research*, **19**, 55-70 (1992)

Eliel *et al.*, *J. Am. Chem. Soc.* **94**(1), 171-176 (1972)

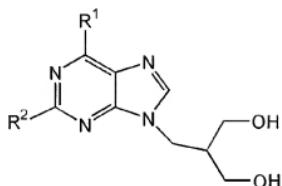
Dunn *et al.*, *J. Org. Chem.* **55**, 6368-6373 (1990)

20 Ludwig *et al.*, *J. Org. Chem.* **54**, 631-635 (1989)

THE CLAIMS

1. Use of a compound of formula (1)

5



10

(1)

where:

R¹ is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or NRCOR';

R² is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or NRCOR'; and

R and R' are independently selected from hydrogen, alkyl and aryl;

or a salt and pharmaceutically acceptable derivatives thereof;

in the treatment and/or prophylaxis of hepatitis B viral infection.

20

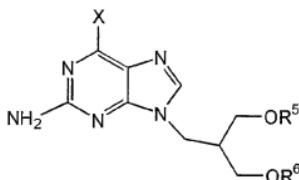
2. Use according to claim 1 wherein R¹ is hydroxy or a group capable of being converted *in vivo* to hydroxy.

- 47 -

3. Use according to claim 1 or claim 2 wherein R² is amino or a group capable of being converted *in vivo* to amino.

4. Use according to any one of the claim 1 to 3 wherein the compound of formula

5 (1), or salt or pharmaceutically acceptable derivative thereof is a compound of formula (4)



(4)

where:

10 X is hydrogen or hydroxy;

R⁵ and R⁶ are the same or different and together with the oxygen atom to which they are attached form a hydroxy group, an ester, a carbonate, a carbamate, or a thiocarbonate;

a salt thereof.

15

5. Use according to claim 4 wherein R⁵ and R⁶ are the same or different and together with the oxygen atom to which they are attached to form a hydroxy group or an ester.

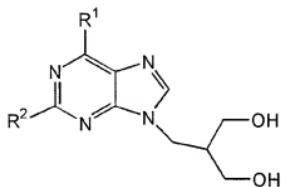
6. Use according to claim 1 wherein R¹ is hydroxy and R² is amino.

20

- 48 -

7. A method for the treatment or prophylaxis of hepatitis B to viral infection which method includes administering to a patient in need thereof an effective amount of a compound of formula (1)

5



10

(1)

where:

R^1 is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or $NRCOR'$;

15 R^2 is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or $NRCOR'$; and

R and R' are independently selected from hydrogen, alkyl and aryl;

or a salt or pharmaceutically acceptable derivative thereof.

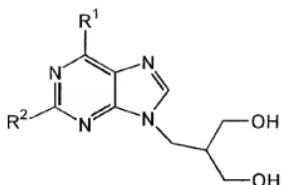
20

8. Use of a compound of formula (1)

25

- 49 -

5



(1)

10 where R¹ is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or NRCOR';

R² is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or NRCOR'; and

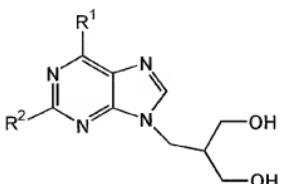
15 R and R' are independently selected from hydrogen, alkyl and aryl;

or a salt or pharmaceutically acceptable derivative thereof;

in the manufacture of a medicament for the treatment or prophylaxis of hepatitis B viral infection.

20 9. A pharmaceutical composition for the treatment or prophylaxis of hepatitis B viral infection including a compound of formula (1)

25



- 50 -

(1)

where:

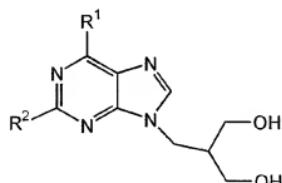
R¹ is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or NRCOR';

5 R² is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or NRCOR'; and

R and R' are independently selected from hydrogen, alkyl and aryl;
or a salt or pharmaceutically acceptable derivative thereof;
10 in association with a pharmaceutically acceptable carrier or diluent.

10. A compound of formula (1a)

15



1a

where:

R¹ is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or NRCOR';
20

- 51 -

R^2 is hydrogen, halogen, hydroxy, azido, alkoxy, aryloxy, thio, alkylthio, amino, alkylamino, hydrazino, hydroxylamino, benzyloxy, NRR' or $NRCOR'$; and

R and R' are independently selected from hydrogen, alkyl and aryl;

or a salt or pharmaceutically acceptable derivative thereof;

5 provided that the following compounds are excluded;

9-[3-hydroxy-2-hydroxymethylprop-1-yl]-guanine, 9-[3-hydroxy-2-hydroxymethylprop-1-yl]-adenine, 9-[(2-isopropyl-1,3-dioxan-5-yl)methyl]-guanine, and 9-[(2-isopropyl-1,3-dioxan-5-yl)methyl]-adenine.

10 11. A compound according to claim 10 which is 2-amino-9-[3-hydroxy-2-hydroxymethylprop-1-yl]purine, 9-[3-acetoxy-2-acetoxymethylprop-1-yl]-2-aminopurine, or a salt or pharmaceutically acceptable derivative thereof.

15 12. A pharmaceutical composition including a compound of formula (1a) as claimed in claim 10 in association with a pharmaceutically acceptable carrier or diluent.

13. A method according to claim 7 wherein the compound of formula (1) or salt or derivative thereof is administered in combination with another pharmaceutical used in the treatment of viral infections.

20

14. A method according to claim 7 wherein administration is via the oral route.

INTERNATIONAL SEARCH REPORT

International application No.

PCT/AU 98/00748

A. CLASSIFICATION OF SUBJECT MATTER		
Int Cl ⁶ : C07D 473/18, 473/32, 473/16, 473/40, 473/24, 473/00 A61K 31/52		
According to International Patent Classification (IPC) or to both national classification and IPC		
B. FIELDS SEARCHED		
Minimum documentation searched (classification system followed by classification symbols)		
Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched		
Electronic data base consulted during the international search (name of data base and, where practicable, search terms used) CHEM. ABS. SUBSTRUCTURE		
C. DOCUMENTS CONSIDERED TO BE RELEVANT		
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	Chemical Abstracts Vol. 123(20), Abstract No. 266118, 13 November 1995 ASHTON, P. et al.: "Codrugs as a method of controlled drug delivery". See Abstract CAS RN 169046-90-6 CAS RN 169046-89-3 & WO 95/20567 (see in particular pages 10-11)	1-10, 12-14
<input checked="" type="checkbox"/> Further documents are listed in the continuation of Box C		<input checked="" type="checkbox"/> See patent family annex
* "A"	Special categories of cited documents: document defining the general state of the art which is not considered to be of particular relevance	"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
* "E"	earlier application or patent but published on or after the international filing date	"X" document of particular relevance, the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
* "L"	document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	"Y" document of particular relevance, the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
* "O"	document referring to an oral disclosure, use, exhibition or other means	"&" document member of the same patent family
* "P"	document published prior to the international filing date but later than the priority date claimed	
Date of the actual completion of the international search 22 October 1998		Date of mailing of the international search report 29 OCT 1998
Name and mailing address of the ISA/AU AUSTRALIAN PATENT OFFICE PO BOX 200 WODEN ACT 2606 AUSTRALIA Facsimile No. (02) 6285 3929		Authorized officer G. D. HEARDER Telephone No.: (02) 6283 2553

INTERNATIONAL SEARCH REPORT

International application No.

PCT/AU 98/00748

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT	
Category*	Citation of document, with indication, where appropriate, of the relevant passages Relevant to claim No.
X	Chemical Abstracts Vol. 109(21), Abstract No. 190136, 21 November 1988 WEBB, R.R. et al.; "Antiviral phosphonomethoxyalkylpurines and -pyrimidines and their preparation". See abstract CAS RN 117087-05-5 CAS RN 117087-06-6 & EP 269947 A1 (see in particular pages 22-23) 1-3, 6-10, 12-14
X	Chemical Abstracts Vol. 115(1), Abstract No. 3301, 8 July 1991, RODRIGUEZ, L. et al.; "Pyrophosphate formation as the most efficient condensation reaction of activated nucleotides", See Abstract CAS RN 121712-76-3 & J. Mol. Evol. (1991), 32(2), 101-4 9, 10, 12
X	Chemical Abstracts Vol. 113(21), Abstract No. 191849, 19 November 1990, BENNER, S.A.; "Isoteric oligonucleotide analogs containing sulfur", See Abstract CAS RN 128435-48-3 CAS RN 128435-49-4 CAS RN 128435-50-7 CAS RN 128435-51-8 & WO 89/12060 A1 9, 10, 12
X	Chemical Abstracts Vol. 112(25), Abstract No. 231404, 18 June 1990, TOHIDI, M. et al.; "Polymerization of the cyclic pyrophosphates of nucleosides and their analogs", See Abstract CAS RN 121712-76-3 CAS RN 121712-77-4 CAS RN 127293-87-2 CAS RN 127317-08-2 & J. Mol. Evol. (1990), 30(2), 97-103 9, 10, 12
X	Chemical Abstracts Vol. 111(7), Abstract No. 52604, 14 August 1989, TOHIDI, M. et al.; "Some acyclic analogs of nucleotides and their template-directed reactions", See Abstract CAS RN 121712-73-0 CAS RN 121712-78-5 CAS RN 121712-84-3 CAS RN 121712-76-3 CAS RN 121712-79-6 CAS RN 121712-85-4 CAS RN 121712-77-4 CAS RN 121712 80-9 CAS RN 121712-86-5 CAS RN 121712-88-7 CAS RN 121712-81-0 CAS RN 121712-90-1 CAS RN 121712-89-8 CAS RN 121712-83-2 CAS RN 121712-91-2 & J. Mol. Evol. (1989), 28(5), 367-73 9, 10, 12

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No.
PCT/AU 98/00748

This Annex lists the known "A" publication level patent family members relating to the patent documents cited in the above-mentioned international search report. The Australian Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

Patent Document Cited in Search Report				Patent Family Member			
EP	269947	AU	81250/87	CA	1339780	DK	6040/87
		EG	19346	IL	84477	JP	63170388
		NZ	222553	PT	86156	ZA	8708607
		US	5650510				
WO	9520567	AU	62545/94	CA	2182228	EP	740650
WO	8912060	AU	37654/89	EP	418309	US	5216141
END OF ANNEX							